Report for 2003CT23B: Occurrence and Fate of Pharmaceuticals in the Pomperaug River

There are no reported publications resulting from this project.

Report Follows

Statement of Critical Regional or Water Problem:

Pharmaceuticals and other compounds of wastewater origin have been observed throughout the US in surface waters impacted by urban activities. The presence of pharmaceuticals in aquatic environments is of concern because, even at ng/L levels, these molecules are biologically active and can affect critical development stages and endocrine systems of aquatic organisms. Current pharmaceutical fate studies have been survey-oriented, only documenting occurrence in a variety of environmental systems. Few data regarding temporal and spatial distributions, or environmental degradation rates of pharmaceuticals in surface waters have been collected. No such studies have been conducted to date in the US.

Environmental occurrence of pharmaceuticals is of particular concern in the Pomperaug River watershed. Here the primary source of pharmaceuticals inputs is a wastewater treatment plant that serves a retirement community of 5000 with an average of 6 medications per person. The treatment plant provides up to 20% of river flow and thus pharmaceutical impacts are expected to be greater in this watershed than the national average.

The objectives of this proposed study are to monitor the temporal and spatial distributions of pharmaceutical compounds introduced to the environment from a well-defined wastewater treatment plant discharge to a river. The specific tasks will include quarterly sample collection from the treatment plant influent and effluent, and at discrete locations downstream in the river channel. Observed concentrations in the river will be compared to predicted concentrations using a conservative transport model to: (1) identify pharmaceutical compounds with potential for ecotoxicological risk in this watershed, and (2) to estimate the magnitude of sink mechanisms for unconserved compounds.

Methods, Procedures and Facilities:

Pharmaceutical Compounds

The choice of pharmaceutical compounds to monitor must take into consideration usage and metabolism rates, the potential for toxicological impacts on non-target organisms and availability of analytical methods suitable to environmental samples. Nation-wide, the top 25 most prescribed drugs include lipid regulators, antihypertensives, hormone replacement therapies, beta-blocker and calcium channel blocker medications and anti-inflammatories [19]. All of these medications are typical of those expected to be used in a senior population.

Of the most widely prescribed drugs, the classes that are known, or strongly suspected, to have ecotoxicological risk are serotonin reuptake inhibitors (*e.g.*, fluoxetine) and beta-blockers (*e.g.*, fenfluramine). Direct toxicological evidence for induced spawning behavior in clams and feminization of crabs has been documented for fenfluramine and fluoxetine (Prozac), respectively, at environmentally relevant concentrations [1]. These induced behaviors arise because of common modes of drug action between humans and these non-target organisms [1].

Finally, the combined analytical advances in solid phase extraction techniques and mass spectrometry detection have pushed detection limits of over 125 pharmaceutical compounds below 0.05 μg/L in environmental samples [17, 18].

On the basis of usage, toxicity and our expertise with GC/MS techniques (see Analysis below), we will monitor estrogens, neutral pharmaceuticals (includes common lipid regulators clofibrate and fenofibrate, pschiatric drug diazepam, β-blockers metoprolol and terbutalin), and acidic drugs (includes anti-imflammatoryies naproxen, diclofenac). Additional pharmaceuticals that will also be isolated with the target analytes can be found in methods of Koplin *et al.* [17] and Ternes [18], and are omitted here for brevity. Usage information that is specific to the Southbury community, including Heritage Village, will be obtained from dispensing records from the local pharmacy and used to tune the final list of analytes.

Sampling Locations and Protocol

Quarterly sample collection will be conducted at the Heritage Water Treatment Plant and through the downstream reach of the Pomperaug River. The proposed sample dates are May, August, November 2003 and February 2004 to encompass relevant seasonal variations.

Heritage Water Treatment Plant: HWTP operates as a conventional activated sludge plant. Influent is coarsely screened, metered and then flows into one of four extended aeration tanks for removal of organic matter and nitrogen. Aeration is followed by clarification to remove biological solids. The clarified water is next chlorinated to deactivate pathogens. This step signifies the end of engineered treatments. The post-chlorination effluent is directed to one of two man-made ponds for dechlorination. These ponds operate as natural systems with no engineered controls. Finally, flow from these ponds discharges by open channel to the Pomperaug River. Average daily capacity of this plant is 0.4 MGPD.

Three sample points have been identified at the Heritage Water Treatment Plant: (1) influent, (2) post-chlorination, and (3) entry to the Pomperaug River. The intermediate sample location between the chlorination treatment and the dechlorination ponds was included to compare pharmaceutical removals in the engineered system with the natural pond system, and ultimately, with losses in the river system. Oral agreement to sample at this facility has been obtained at the time of this proposal submission (Ray Adamaitis, CT Water Company, personal communication).

Pomperaug River: The Pomperaug River is a third-order stream with typical median flows that range from 10 – 30 cfs, as measured at a USGS gauging station 890 m (0.56 mi) downstream of the HWTP discharge. The river reach between HWTP and the Housatonic River is 9700 m (6.1 mi) with only 2 minor brook inputs. Throughout this reach, the river channel is wadeable with a rocky bottom and steep banks, as is characteristic of Connecticut rift geology.

Samples will be collected for pharmaceutical analysis at 5 locations downstream from the HWTP discharge point (Figure 1). One sampling point will be coincident with the USGS gauging station where the most accurate calculations of relative wastewater and channel flow contributions to the Pomperaug River can be made. The other sample points are spaced approximately to a logarithmic scale to improve estimates of environmental degradation rates for the monitored analytes. All of the sample locations are bordered by properties that are town- or land trust-owned, and thus are accessible by the public. Sample locations may be modified to better capture dynamics of pharmaceutical concentrations based upon results of the May 2003 sampling event.

The HWTP has been identified as the sole point source of pharmaceutical compounds in the Pomperaug River. However, the high use of septic treatment systems throughout the

watershed indicate that non-point sources of pharmaceuticals may also be introduced to the Pomperaug River through groundwater flows. Thus, a sixth river water sample will be collected 500 m upstream of the HWTP discharge point to assess background concentrations of pharmaceuticals that are transported into the study area by Pomperaug River flow.

Samples will be collected on a flow-corrected time schedule so that a single 'packet' of water is followed from the HWTP discharge. This technique will enable conversion of spatial variations in pharmaceutical concentrations to reaction time equivalents in a batch reactor, and hence meaningful estimates of pharmaceutical attenuation rates will be possible. Standard stream tracer techniques will be employed to delineate the fluid 'packet' using fluorescent rhodamine tracer [20]. To further verify channel dilution effects due to infiltration of groundwater, boron and chloride concentrations will be measured at the sample points. Boron is a common tracer of wastewater effluents [21]. Chloride concentrations in wastewater [50 – 90 mg/L, 21] are greater than in freshwater streams [~ 2 mg/L, 22], although other sources (*e.g.*, manure leachate) may contribute chloride to the Pomperaug River.

Samples for pharmaceutical analyses will be collected in clean 1-L polypropylene sample bottles. A total of 4 samples will be collected by immersion at each location. An additional 250 mL sample will be collected for analysis of conserved wastewater tracers in the Pomperaug River. Samples will be stored in a cooler for transport back to Storrs. Samples will be preserved by refrigeration at 4°C and extracted within 72 hours. A unique sample ID number will be assigned to each sample. All treatments will be documented in a bound log-book.

Where possible, river sediment grab samples will also be collected at the sample points during the November 2003 sampling trip. These samples will be extracted to give qualitative verification of whether sediments are important sinks for pharmaceuticals in this watershed. Sediment samples will be collected with a clean metal trowel, transferred to a polypropylene sample bottle and preserved by freezing (-20°C) until analysis.

Analytical

Pharmaceutical analyses in water samples will be conducted using published methods [17, 18]. The four 1-L samples from each location will be compositted and then split to quantify estrogenic pharmaceuticals, neutral pharmaceuticals and acidic pharmaceuticals. Appropriate recovery standards will be added to each sample to account for compound losses in sample analysis methods. Briefly, the method for each of the compound classes entails: (1) pharmaceutical extraction by passage through a solid-phase extraction column; (2) elution with appropriate solvent, and (3) addition of a derivatizing agent to increase pharmaceutical volatility. Slight differences in water pre-treatments, solvent types and derivatization conditions necessitate the use of 3 parallel analyses. Derivatized products will be quantified by gas chromatography (GC) with mass spectrometry (MS) detection (Shimadzu QP5050A GC/MS system available in PI's lab). If interferences from wastewater constituents limit GC/MS detection for certain key samples, liquid chromatography (LC) with MS detection will be applied for quantification. LC/MS analysis is available from the Chemistry Department at UConn on a fee-per-sample basis.

A number of quality assurance/quality control (QA/QC) checks will be incorporated into the sample design. First, each sample trip will include trip blanks consisting of high purity water that is transferred to a sample container and transported to the sample site. Method blanks

consisting of high purity water will be carried through all of the sample preparation and analysis steps. Method spikes will be prepared by adding pharmaceutical compounds to high purity water or samples of wastewater effluent and analyzed to assess compound recoveries. QA/QC checks will account for 10% of sample analyses with a minimum of one trip blank, one method blank and one method spike included in each sampling round. Prior to each sample trip, 2 of the 9 sample locations will be chosen randomly for obtaining duplicate samples.

Boron analyses will be conducted by inductively coupled plasma (ICP) spectrometry (for fee service available from Environmental Research Institute, UConn) and chloride concentrations will be quantified using an ion selective electrode.

Sediment samples will be analyzed using extraction techniques that are currently under development in our lab as part of a USDA-funded investigation of antibiotic fates in agricultural soils. Present sediment extraction techniques (*e.g.* EPA Method 3550) yield inadequate recoveries of polar pharmaceutical compounds; however, competitive displacement, pH and ionic strength adjustments are promising alternate approaches (MacKay, unpublished results).

In all sample analysis techniques, instruments will be calibrated with a 6-point calibration curve. Calibration standards will be run as unknowns every 8 analyses with acceptance criteria of ± 15%, or as determined in method development.

Conceptual Modeling

Ultimately, the results obtained in this study will be used to assess environmental degradation rates of pharmaceutical compounds. First, observed pharmaceutical concentrations in the Pomperaug River will be compared to expected concentrations given dilution effects of inflowing groundwater and minor stream contributions to the river channel. Dilution effects will be calculated from the stream tracer measurements using the added fluorescent tracer (dispersive mixing of sample packet) and the wastewater-derived boron and chloride tracers (fractional contribution of wastewater to total flow) using standard techniques [20]. Insignificant differences on a compound-by-compound basis between observed fluxes and calculated dilution-corrected pharmaceutical fluxes at each downstream sampling point indicate pharmaceutical compounds that undergo no transformations or have no significant loss mechanisms within this ecosystem. On the other hand, observed pharmaceutical fluxes that are lower than obtained solely by dilution will indicate that sinks for these compounds are important within the study reach.

Characteristic environmental degradation rates will be estimated for unconserved pharmaceutical compounds using a simple first-order loss model [20]. This quantitative approach will yield the first reported environmental degradation rate constants for compounds in the pharmaceutical class. However, this approach gives little insight into the exact mechanisms of loss since it quantifies the summative effect of multiple processes (*e.g.*, photodegradation, microbial degradation, sorptive uptake by sediments).

Work Plan and Time Line

PI Dr Allison MacKay will oversee this research project, providing guidance for analytical methods and QA/QC protocols. Sample collection and analysis, including method validation, will be conducted by graduate research assistant Ms. Raquel Figueroa. The following timeline marks milestones in project completion:

March 2003 – Method assessment and validation with genuine standards, preliminary dilution ratio characterization in river channel.

May 2003 – Spring sample collection and data analysis.

August 2003 – Summer sample collection and data analysis.

November 2003 – Fall sample collection and data analysis.

February 2004 – Winter sample collection and data analysis.

March 2004 – Presentation of results to Pomperaug River Watershed Coalition and manuscript preparation.

Significant Findings and Results:

We have requested a no-cost extension for CTIWR "Fate of Pharmaceuticals in the Pomperaug River" until Dec. 31, 2004. A no-cost extension is requested because field activities proposed in this grant could not be completed due to the unexpectedly high levels of precipitation in CT during Summer and Fall 2003. We intend to complete these activities in Summer and Fall 2004.